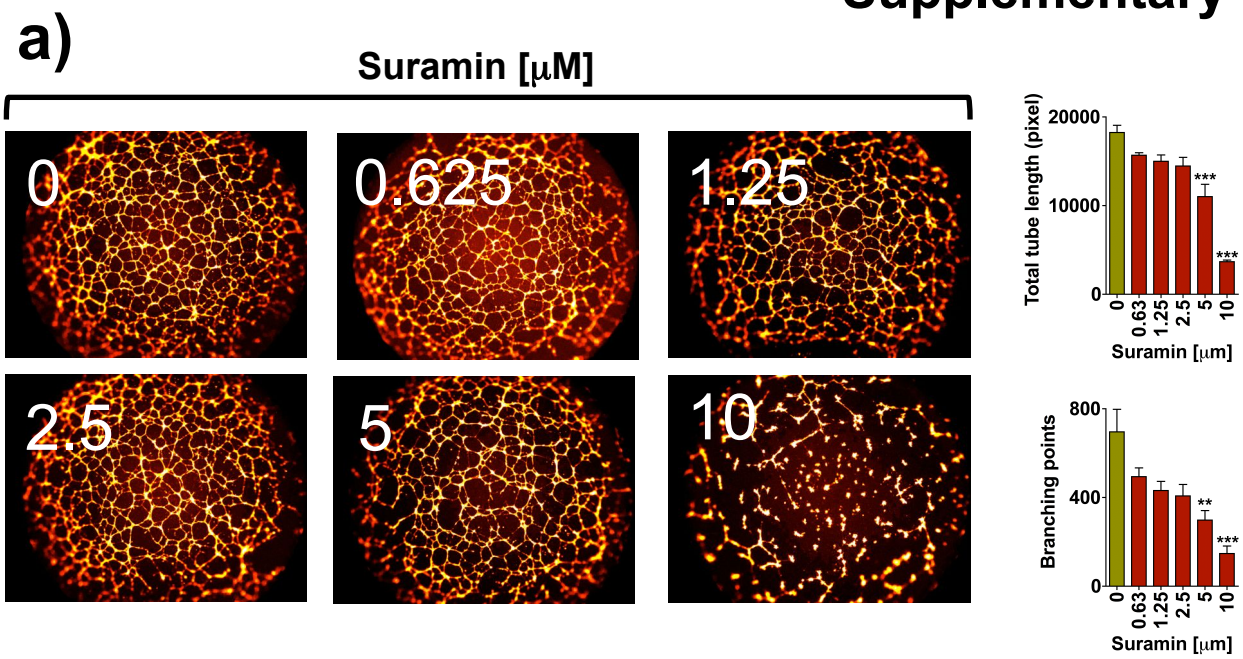
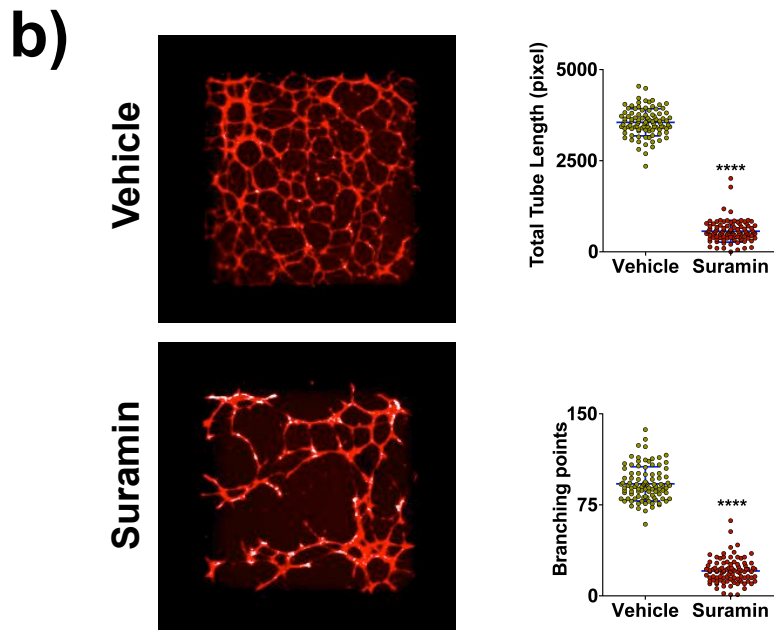
Supplementary Fig 1





Supplementary Figure 1. Miniaturization and automation of Endothelial tube formation assay. **a)** 96-well plate format: HUVEC (15,000 cells/100μl media) were seeded on top of matrigel (50μl) and incubated at 37°C/CO_2 %5 for 8 hours. Endothelial tubes stained with Phalloidin alexa 568 and imaged at 2x objective using an Operetta high content imaging system. Suramin was used as positive control. DMSO (vehicle) was used as negative control. Two parameters: total tube length and branching points were analyzed using Metamorph image analysis software, n=3 wells per concentration. Error bars, mean±s.e.m, one-way ANOVA followed by Bonferroni's *post hoc* test, **p<0.01; ***p<0.001 compared to vehicle. **b)** 384-well plate format: HUVEC (4,500 cells/32μl media) were seeded on top matrigel (16μl), Staining and imaging procedures were performed as described above, n=96 wells per condition. Data are presented as scatter plot with mean±S.D. Unpaired-*t*-test, ****p<0.0001 compared to vehicle. Z' factor was calculated using vehicle and Suramin (8μM) treated wells. Z' factor (Total tube length)=0.33 and Z' factor (branching points)<0.3.

Supplementary Fig 2

a)

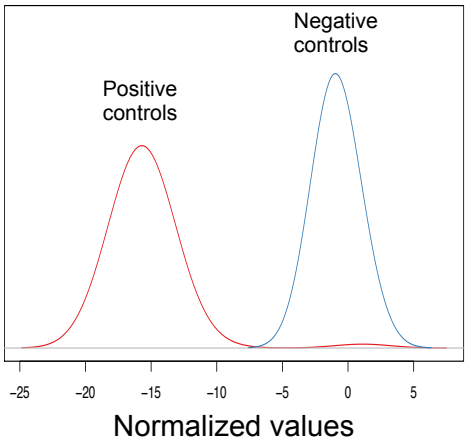
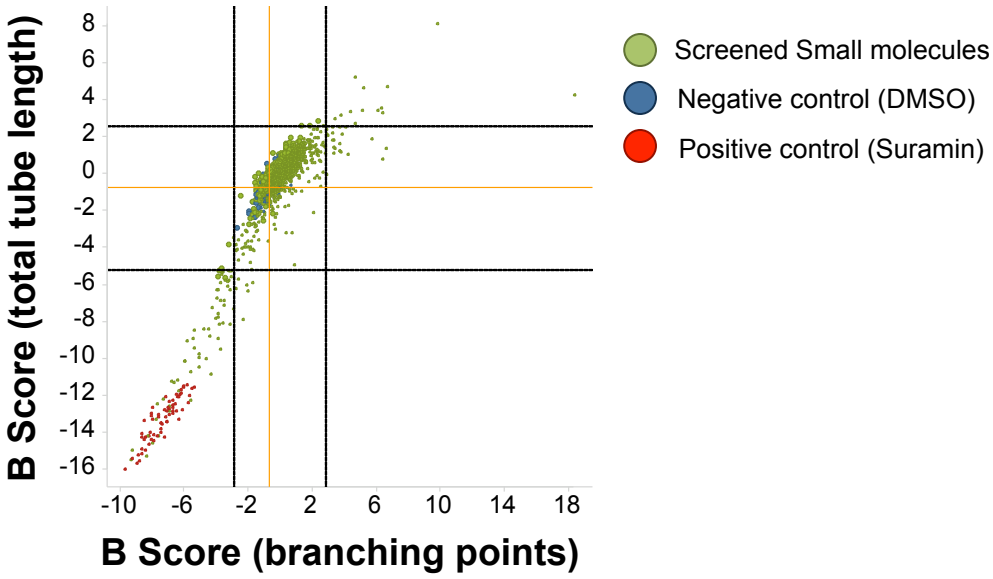


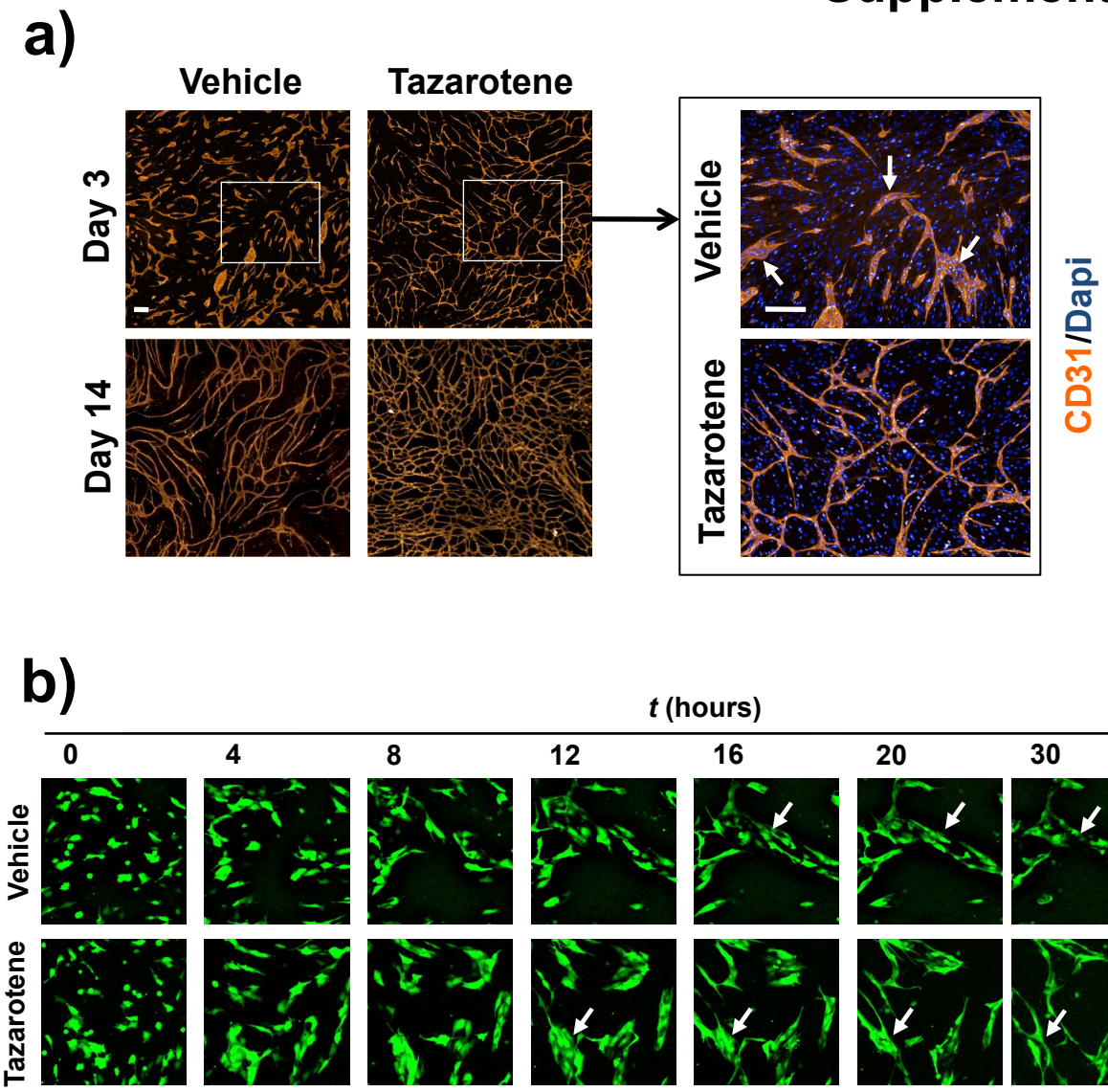
Plate	*Replicate*	*Z' factor*
1	1	0.368
1	2	0.575
2	1	0.379
2	2	0.638
3	1	0.312
3	2	0.483
4	1	0.658
4	2	0.488

b)



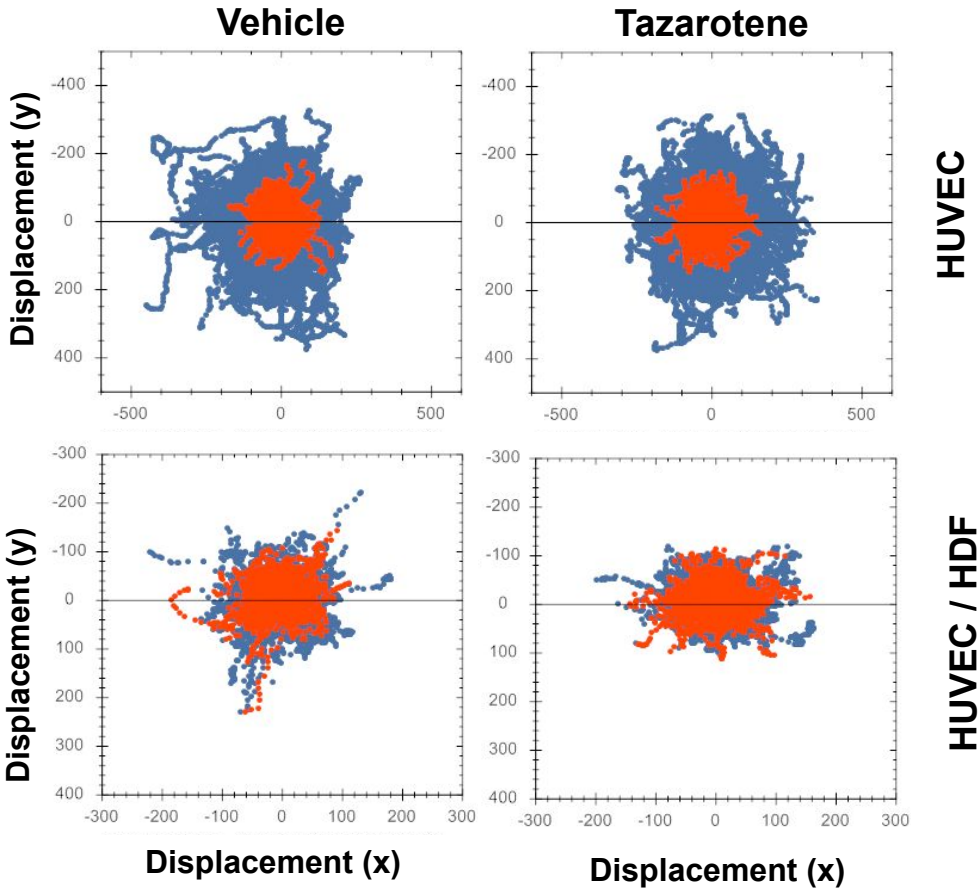
Supplementary Figure 2. a) Quality control for screen plates was evaluated using standard Z' factor values for every screen plate. **b)** Scatter plot of data screen showing the positive correlation between total tube length and branching points, r^2=0.85.

Supplementary Fig 3



Supplementary Figure 3. Effect of Tazarotene (3μM) on the tubule remodeling. **a)** Representative images showing the early stage (3 days) and the established stage (14 days) of tube formation following the co-culture of HUVEC and fibroblasts (HDF), n=6 wells per condition. HUVEC were stained with anti-CD31 antibody (orange) and Nuclei with DAPI (blue). Arrows indicate endothelial cell clusters. Scale bar 200 μm. **b)** HUVEC expressing eGFP were seeded onto confluent fibroblast (HDF) in 96-well plate and tube formation was followed by time-lapse live cell imaging using the high-content imaging system (Operetta). Images were captured every 15 min over 48 hours using 10x objective, (**Supplementary Movies 1&2**). Video stills are shown at 4 hour intervals over a 40 hour period for the early stage of tubulogenesis. 2 fields per well, n=3 wells per condition. Arrow refers to the remodeling process of endothelial cluster into tube.

Supplementary Fig 4



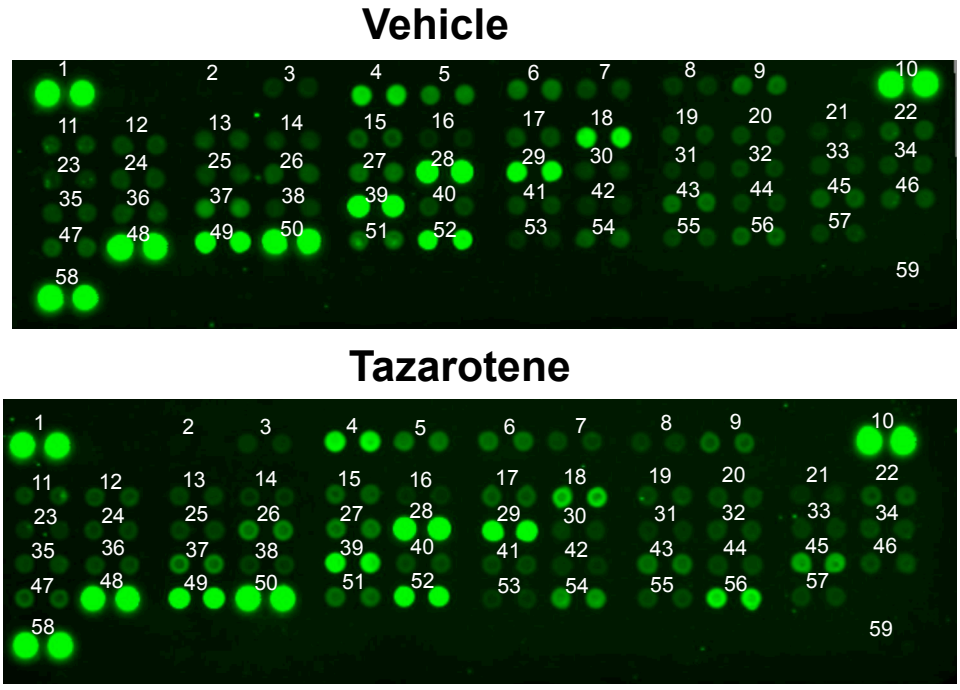
Supplementary Figure 4. Effect of Tazarotene on endothelial cell motility. HUVEC expressing eGFP were cultured in 96-well plate as single cell culture or co-cultured with fibroblasts (HDF). Images were captured every 15 min over 24 hours using 10x objective. 2 fields per well. Cells were identified and tracked over time using Harmony Software. Cell displacement was calculated for more than 200 cells per well. Every dot represents the cell position. Red dots the cell tracks during the first 6 hours, n=3 wells per condition.

Supplementary Fig 5

a)

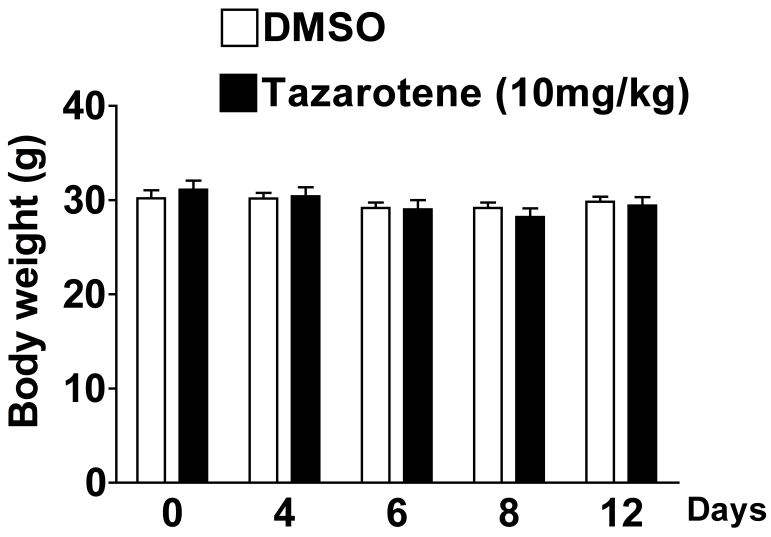
Index	Target/Control
1	Reference Spots
2	Activin A
3	ADAMTS-1
4	Angiogenin
5	Angiopoietin-1
6	Angiopoietin-2
7	Angiostatin/Plasminogen
8	Amphiregulin
9	Artemin
10	Reference Spots
11	Coagulation Factor III
12	CXCL16
13	DPPIV
14	EGF
15	EG-VEGF
16	Endoglin
17	Endostatin/Collagen XVIII
18	Endothelin-1
19	FGF acidic (FGF1)
20	FGF basic (FGF2)
21	FGF-4
22	FGF-7
23	GDNF
24	GM-CSF
25	HB-EGF
26	HGF
27	IGFBP-1
28	IGFBP-2
29	IGFBP-3
30	IL-1beta
31	IL-8
32	LAP (TGFbeta1)
33	Leptin
34	MCP-1
35	MIP-1alpha
36	MMP-8
37	MMP-9
38	NRG1-beta1
39	Pentraxin
40	PD-ECGF
41	PDGF-AA
42	PDGF-AB/PDGF-BB
43	Persephin
44	Platelet Factor 4
45	PIGF
46	Prolactin
47	Serpin B5
48	Serpin E1
49	Serpin F1
50	TIMP-1
51	TIMP-4
52	Thrombospondin-1
53	Thrombospondin-2
54	uPA
55	Vasohibin
56	VEGF
57	VEGF-C
58	Reference Spots
59	Negative Control

b)



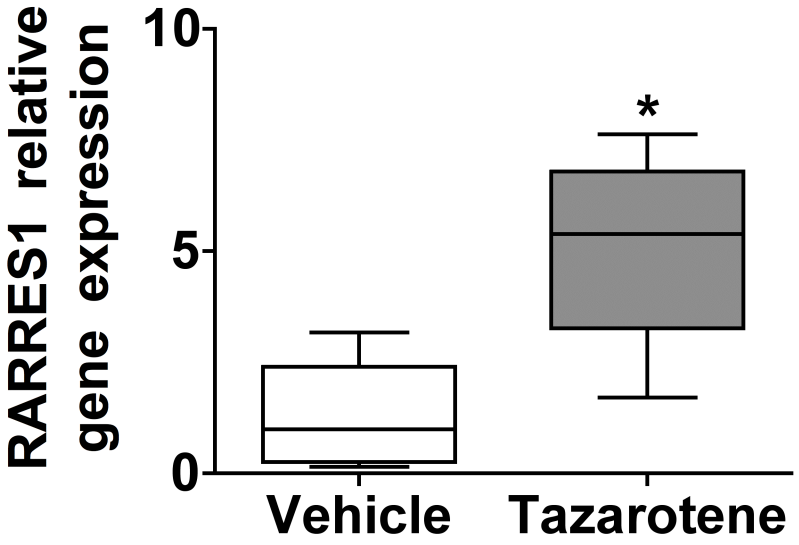
Supplementary Figure 5. Effect of Tazarotene on angiogenesis-related secreted proteins from human dermal fibroblasts using Proteome Profiler Array™. a) the table showing the human angiogenesis cytokines present in the array. **b)** Raw blots showing the array membrane for Vehicle (top) and Tazarotene (below), with numbers corresponding to those on the table.

Supplementary Fig 6



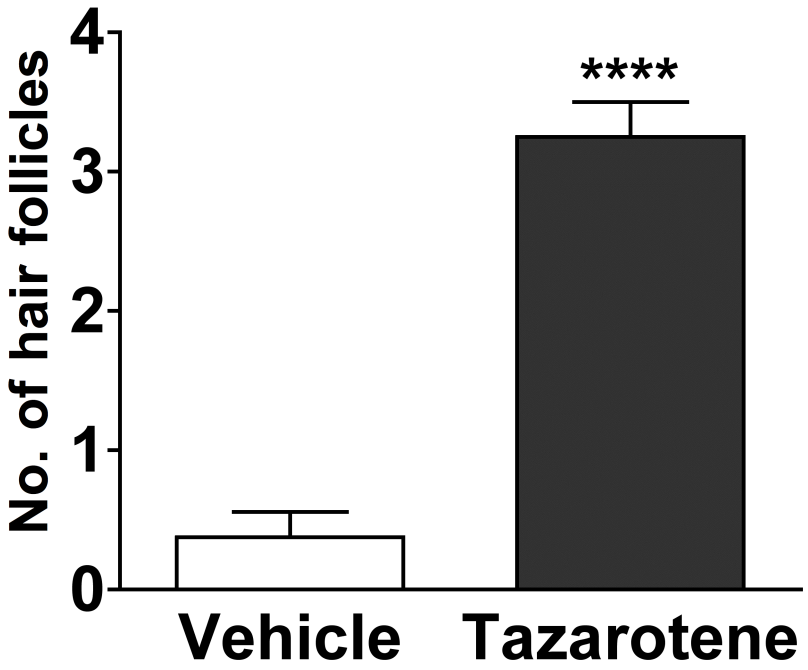
Supplementary Figure 6. Effect of systemic administration of Tazarotene on the mouse body weight. Tazarotene (10mg/kg) was injected i.p. daily for the first six days. Data are expressed as mean±s.e.m, n=6 animals per group.

Supplementary Fig 7



Supplementary Figure 7. Effect of injected Tazarotene on RARRES1 mRNA level in the liver. Tazarotene (10mg/kg) was injected i.p. in adult mice (8 week old). Mice killed and liver was harvested for RNA analysis 22 hours after injection. Total RNA was extracted for performing quantitative RT-PCR using specific primers for RARRES1, n=4 animals per group. Data expressed as box plot with maximum, minimum and quartile range. Unpaired-*t*-test, *p<0.05 compared to vehicle group.

Supplementary Fig 8



Supplementary Figure 8. Effect of systemic administration of Tazarotene (10mg/kg/day) for 6 days on the growth of new hair follicles in the regenerated area of ear wounds. On H&E stained sections, hair follicles were counted inside the regenerated area which is identified by the cartilage cut, n=4 animals per treatment condition. Data are expressed as mean ±s.e.m. Unpaired-*t*-test, ****p<0.0001 compared to vehicle group.

Supplementary Table 1

Compound name	B Score (Total Tube length)	Class	Selectivity	MW	Description
IC 261	-15.5	Phosphorylation	CK-1delta/epsilon	311.3	Casein kinase-1 (CK-1delta/epsilon) inhibitor.
Iodoacetamide	-15.3	Biochemistry		185.0	Alkylating reagent for cysteine and histidine residues in proteins; irreversible protein inhibitor
Vinblastine sulfate salt	-15.0	Cytoskeleton and ECM	Tubulin	909.1	Inhibitor of microtubule assembly
Stattic	-14.2	Gene Regulation	STAT3	211.2	Irreversible STAT3 activation inhibitor.
Ivermectin	-14.2	Cholinergic	alpha7 nACh	875.1	Positive allosteric modulator of alpha7 neuronal nicotinic acetylcholine receptor; also modulates glutamate-GABA-activated chloride channels
Rotenone	-13.3	Cell Stress	Mitochondria	394.4	Inhibitor of mitochondrial electron transport
Sanguinarine chloride	-12.9	Ion Pump	Na+/K+ ATPase	367.8	Inhibitor of Mg2+ and Na+/K+-ATPase; isolated from the leaves and stems of Macleaya cordata and microcarpa
Bay 11-7085	-12.6	Cell Cycle	IkB-alpha	249.3	Inhibits cytokine induced IkB (Inhibitor of NFkB) phosphorylation
Aurothioglucose	-12.5	Phosphorylation	rac1	392.2	Potent PKC iota - Par6 interaction inhibitor; Disrupts a rac1 signaling pathway required for growth of a cancer cell line.
NSC 95397	-12.3	Phosphorylation	Cdc25	310.4	Selective, irreversible Cdc25 dual specificity phosphatase inhibitor.
Vincristine sulfate	-12.2	Cytoskeleton and ECM	Tubulin	923.1	Inhibitor of microtubule assembly
7-Chloro-4-hydroxy-2-phenyl-1,8-naphthyridi	-11.8	Adenosine	A1	256.7	A1 adenosine receptor antagonist
Pifithrin-mu	-11.3	Apoptosis	p53	181.2	Anti-apoptotic; inhibits p53 binding to mitochodria as well as Bcl-xL and Bcl-2 proteins. Does not affect transcriptional functions of p53.
cis-4-Aminocrotonic acid	-11.2	GABA	GABA-C	101.1	GABA-C receptor agonist
Colchicine	-11.2	Cytoskeleton and ECM	Tubulin	399.4	Prevents tubulin polymerization
Bay 11-7082	-10.9	Phosphorylation	IkB-alpha	207.3	Inhibitor of cytokine-induced IKB-alpha phosphorylation
Terfenadine	-10.5	Histamine	H1	471.7	Non-sedating H1 histamine receptor antagonist
2-methoxyestradiol	-10.2	Hormone	Estrogen	302.4	Inhibitor of angiogenesis and endothelial cell proliferation
5HPP-33	-10.1	Cytoskeleton and extracellular matrix	microtubules	323.4	5HPP-33 is a thalidomide derivative which was reported to be active as anticancer agent through stabilization of the microtubules
JS-K	-10.1	Nitric Oxide		384.3	Nitric oxide donor; antiproliferative
N-p-Tosyl-L-phenylalanine chloromethyl keto	-9.9	Biochemistry	Chymotrypsin alpha	351.9	Blocks LPS- or cytokine-induced activation of nuclear factor kB (NFkB), which, blocks the induction of iNOS and COX-2 transcription; blocks activation of pp70s6k by all mitogens
Z-L-Phe chloromethyl ketone	-9.5	Biochemistry	Chymotrypsin A-gamma	331.8	Bovine chymotrypsin A-gamma inhibitor
Podophyllotoxin	-9.5	Cytoskeleton and ECM	microtubules	414.4	Antineoplastic glucoside; inhibitor of microtubule assembly
PD-166285 hydrate	-9.0	Kinase/Phosphatase	Src, FGFR	603.4	PD-166285 hydrate is a broad spectrum protein tyrosine kinase inhibitor; Src and FGFR kinase inhibitor
BNTX maleate salt hydrate	-9.0	Opiods	d1	545.6	BNTX is a selective d1 non peptide opioid receptor antagonist. BNTX is used as a tool for the study of various function of opioid receptors including alcohol and drug dependence
PD-180970	-8.9	Cell signaling/ tyrosine kinase	p210 Bcr-Abl	429.3	PD-180970 is a potent inhibitor of the p210 Bcr-Abl tyrosine kinase.
Nocodazole	-8.8	Cytoskeleton and ECM	beta-tubulin	301.3	Disrupts microtubules by binding to beta-tubulin
Diphenyleneiodonium chloride	-8.5	Nitric Oxide	eNOS	314.6	Endothelial nitric oxide synthase inhibitor
Amsacrine hydrochloride	-8.4	DNA Repair	TopoII	429.9	DNA topoisomerase II inhibitor
K114	-8.4	Neurotransmission		393.3	A fluorescent, amyloid-specific dye, an analogue of Congo Red and BSB that recognizes amyloid lesions and allows the quantitative monitoring of the formation of amyloid fibrils assembled from the Aß peptide, a-synuclein, and tau.
IPA-3	-8.2	Phosphorylation	Pak1	350.5	allosteric inhibitor of Pak1, binds to autoinhibitory domain of Pak1 (p21 activated kinase)
Emetine dihydrochloride hydrate	-8.1	Apoptosis		553.6	Apoptosis inducer; RNA-Protein translation inhibitor
Hydroquinone	-8.1	Leukotriene	Arachidonate 12-Lipoxygenase	110.1	Arachidonate 12-Lipoxygenase inhibitor
Wiskostatin	-7.9	Actin	N-WASP	426.2	Wiskostatin is a selective inhibitor of N-WASP, a ubiquitously expressed member of the Wiskott-Aldrich Syndrome protein (WASp) family that regulates actin polymerization
N-Oleoyldopamine	-7.8	Neurotransmission	CB1	417.6	Endogenous vanilloid; weak CB1 cannabinoid receptor ligand.
Supercinnamaldehyde	-7.8	Ion Channels	TRPA1	201.2	Transient receptor potential ankyrin 1 (TRPA1) receptor activator; covalently modifies cysteines to acitvate channel.
Calcimycin	-7.6	Intracellular Calcium	Ca2+	523.6	Ca2+ ionophore used to potentiate responses to NMDA, but not quisqualate glutamate receptors
Piperlongumine	-7.3	Apoptosis	GSTP1	317.3	Piperlongumine selectively kills cancer cells regardless of p53 status without harming normal cells. It binds to and inihbits proteins known to regulate oxidative stress
Brefeldin A from Penicillium brefeldianum	-7.3	Cytoskeleton and ECM	Golgi apparatus	280.4	Fungal metabolite that disrupts the structure and function of the Golgi apparatus
PD173952	-7.1	Kinase	Src	482.4	PD173952 is a Src family kinase inhibitor.
Dihydrooouabain	-6.9	Ion Pump	Na+/K+ Pump	586.7	Sodium-potassium pump inhibitor
Parthenolide	-6.9	Serotonin		248.3	Inhibits serotonin release from platelets; inhibits production of leukotriene B4 and thromboxane B2
Mitoxantrone	-6.8	DNA Metabolism		517.4	DNA synthesis inhibitor
Calmidazolium chloride	-6.4	Intracellular Calcium	Ca2+ATPase	687.7	Potent inhibitor of calmodulin activation of phosphodiesterase; strongly inhibits calmodulin-dependent Ca2+-ATPase
PD-166866	-6.3	Tyrosine Kinase	FGF-1	396.5	PD-166866 is a selective inhibitor of the FGF-1 receptor tyrosine kinase (FGFR1) with IC50 = 55 nM, and no effect on c-Src, PDGFR-b, EGFR or insulin receptor tyrosine kinases or MEK, PKC, and CDK4.
Ouabain	-6.3	Ion Pump	Na+/K+ ATPase	584.7	Blocks movement of the H5 and H6 transmembrane domains of Na+-K+ ATPases
SR 27897 hydrate	-6.3	Cholecystokinin	CCK1	411.9	SR 27897 is a potent, selective antagonist of the Cholecystokinin (CCK) receptor CCK1
Tyrphostin AG 879	-6.2	Phosphorylation	TrkA	316.5	Tyrosine kinase nerve growth factor receptor (TrkA) inhibitor; inhibits 140 trk protooncogene and HER-2
MNS	-6.1	Phosphorylation	Src/Syk	193.2	Src and Syk kinase inhibitor; prevents phosphorylation and cytoskeletal association of GPIIb/IIIa and talin.
Chelerythrine chloride	-6.0	Phosphorylation	PKC	383.8	PKC inhibitor; affects translocation of PKC from cytosol to plasma membrane
PD-161570	-5.9	Tyrosine kinase	FGF-1	532.5	PD-161570 is an inhibitor of human FGF-1 receptor tyrosine kinase.
Retinoic acid p-hydroxyanilide	-5.9	Cell Cycle		391.6	Vitamin A acid analog with antiproliferative activity in cultured human breast cancer cells
N-Phenylanthranilic acid	-5.8	Cl- Channel		213.2	Cl- channel blocker
XCT790	-5.8	Gene Regulation	ERRalpha	596.4	Potent and selective estrogen-related receptor alpha (ERRalpha) inverse agonist.
(S)-(+)-Camptothecin	-5.8	Apoptosis	Topol	348.4	DNA topoisomerase I inhibitor
IMS2186	-5.7	Cell Cycle	G2	296.3	IMS2186 is an anti-proliferative and anti-angiogenic. The proposed mechanism action, under investigation, is blocking the cell cycle at G2 and inhibition of the production of PGE2/TNF-a.
Idarubicin	-5.7	DNA Metabolism		497.5	Antineoplastic
Sunitinib malate	-5.6	Tyrosine Kinase		532.6	Sunitinib malate is a receptor tyrosine kinase inhibitor, which targets VEGF-R1, VEGF-R2, VEGF-R3, PDGF-Ra, PDGF-Rß, KIT, FLT3, CSF-1R, and RET. Sunitinib malate is an anticancer drug.
Moclobemide	-5.6	Neurotransmission	MAO	268.7	Reversible monoamine oxidase A inhibitor (MAOI); antidepressant.
BTO-1	-5.4	Phosphorylation	Plk	264.2	Polo-like kinase (Plk) inhibitor.
PF-431396 hydrate	-5.3	Kinase inhibitors	PYK2 and FAK	506.5	PF-431396 is a potent inhibitor of PYK2 and FAK kinases
TBBz	-5.3	Phosphorylation	CK2	433.7	Cell-permeable casein kinase 2 (CK2) inhibitor.
SU 4312	-5.3	Phosphorylation	KDR	264.3	Vascular endothelial growth factor (VEGF) receptor protein tyrosine kinase 1/2 (KDR) and platelet derived growth factor (PDGF) receptor inhibitor
Topotecan hydrochloride hydrate	-5.2	Apoptosis and Cell Cycle	topoisomerase I	475.9	Topetecan is a topoisomerase I inhibitor and an apoptosis inducer. It is a potent antineoplastic agent
DL-erythro-Dihydrosphingosine	-5.2	Phosphorylation	PKC / PLA2 / PLD	301.5	Protein kinase C, phospholipase A2, and phospholipase D inhibitor
Cilostamide	-5.2	Cyclic Nucleotides	PDE III	342.4	cGMP-inhibited phosphodieserase inhibitor (PDE III)
Auranofin	-4.9	Phosphrylation		678.5	Inhibits the release of inflammatory mediators from human macrophages, basophils, and pulmonary mast cells. Inhibits 5-lipoxygenase in human neutrophils. Is a potent inhibitor of selenoenzyme thioredoxin reductase (TrxR). Auranofin inhibits IKB kin...
Bosutinib	-4.8	kinase	Src/Abl	530.5	Bosutinib (SKI-606) is an orally active; dual Src/Abl tyrosine kinase inhibitor with potent antiproliferative activity.
Pirfenidone	-4.7	Immune System		185.2	Inhibitor of collagen production and fibroblast proliferation
Niclosamide	-4.4	Antibiotic	Protonophore	327.1	Protonophoric anthelmintic; uncouples oxidative phosphorylation
beta-Lapachone	-4.2	Apoptosis		242.3	Induces apoptosis in HL-60 cells; anticancer agent
Trimipramine maleate	-4.2	Serotonin	Reuptake	410.5	Serotonin reuptake inhibitor that also blocks norepinephrine reuptake; antidepressant
PD-407824	-4.1	Kinase	Wee1/Chk1	328.3	PD-407824 is a Wee1/Chk1 inhibitor
6-Nitroso-1,2-benzopyrone	-4.1	Transcription	PARP	175.1	Poly(ADP-ribose) polymerase (PARP) ligand which preferentially destabilizes one of the two zinc-fingers inactivating the enzyme
Debrisoquin sulfate	-4.1	Neurotransmission		448.5	Antihypertensive; metabolized to tetrahydroisoquinoline (TIQ) which may play a role in the induction of Parkinson's Disease
SU 5416	-4.0	Phosphorylation	VEGFR PTK	238.3	Potent and selective VEGFR PTK inhibitor; inhibits VEGF-induced angiogenesis
Ellipticine	-4.0	Cell Cycle	CYP1A1 / TopoII	246.3	Cytochrome P450 (CYP1A1)and DNA topoisomerase II inhibitor
PF-573228	-4.0	Kinase/Phosphatase		491.5	PF-573228 is a focal adhesion kinase (FAK) inhibitor; Non-receptor tyrosine kinase inhibitor
PD 0325901	-4.0	Kinase phosphotase biology	MKK1(MEK1) & MKK2 (MEK2)	482.2	PD 0325901 is a potent MKK1 (MEK1) and MKK2 (MEK2) inhibitor

Supplementary Table 1. Hit list illustrates all the compounds showed reduction of branching and total tube length in the primary high content screening using endothelial tube formation assay. SU 4312 and SU 5416 are inhibitors of VEGF receptors, which can be considered as internal positive controls.

Supplementary Table 2

Compound	*B Score (Total Tube length)*	*Class*	*Selectivity*	*MW*	*Description*
GBR-12909 dihydrochloride	8.1	Dopamine	Reuptake	523.5	Selective dopamine reuptake inhibitor
Tetrabenazine	5.3	Neurotransmission	VMAT	317.4	Reversible type 2 vesicular monoamine transporter (VMAT) inhibitor. It depletes dopamine stores.
Trequinsin hydrochloride	4.7	Cyclic Nucleotides	PDE III	442.0	Phosphodiesterase III (PDE III) inhibitor
GW2974	4.6	Phosphorylation	EGFR / ErbB-2	395.5	Dual EGFR and ErbB-2 receptor tyrosine kinase inhibitor
KB-R7493	3.6	Ca2+ Channel	NCX	427.5	KB-R7943 inhibits the reversed Na(+)/Ca(2+) exchanger, NCX. In cardiomyocytes
1,3,5-tris(4-hydroxyphenyl)-4-propyl-1H-pyrazole	3.5	Hormone	ER-alpha	386.5	Specific estrogen receptor alpha (ERalpha) agonist
Methiothepin mesylate	3.4	Serotonin	5-HT1E, 5-HT1F, 5-HT6	452.7	5-HT1 Serotonin receptor antagonist; blocks serotonin autoreceptors
Y-27632 dihydrochloride	3.4	Phosphorylation	ROCK	320.3	Highly potent, cell-permeable, and selective Rho-associated coiled-coil forming protein serine/threonine kinase (ROCK) inhibitor. Also inhibits ROCK-II with equal potency. The inhibition is competitive with respect to ATP.
LP 12 hydrochloride hydrate	3.3	Serotonin	5-HT7	518.1	5-HT7 receptor agonist displaying selectivity over D2, 5-HT1A and 5-HT2A receptors
Protriptyline hydrochloride	3.2	Adrenoceptor	Reuptake	299.8	Norepinephrine reuptake blocker
Tazarotene	3.0	Cell Biology	RARb, RARg	351.5	Tazarotene is a prodrug of tazarotenic acid, which specifically activates RARb and RARg, only weakly activates RARa. It induces the expression of tazarotene-induced gene 3 (TIG3), a tumor suppressor gene.
Meloxicam sodium	2.9	Prostaglandin	COX-2	373.4	Non-steroidal anti-inflammatory drug (NSAID) that shows 300-fold selectivity for cyclooxygenase-2 (COX-2) vs. cyclooxygenase-1 (COX-1).
SB 202190	2.7	Phosphorylation	p38 MAPK	331.4	Highly selective, potent and cell permeable p38 MAP kinase inhibitor.
N6-2-(4-Aminophenyl)ethyladenosine	2.6	Adenosine	A3	386.4	Potent, non-selective A3 adenosine receptor agonist
Droxinostat	2.6	Gene Regulation	HDAC3, HDAC6, and HDAC8	243.7	Droxinostat is a selective inhibitor of HDAC3, HDAC6, and HDAC8.

Supplementary Table 2. Hit list illustrates all the compounds showed enhancement of total tube length in the primary high content screening using endothelial tube formation assay.